

**Table 1**

*Saprolegnia diclina* (ATCC 56851) Desaturase Expression in Baker's Yeast

CLONE	TYPE OF ENZYME ACTIVITY	% CONVERSION OF SUBSTRATE	
pRSP1 ( <i>S. diclina</i> Δ6 desaturase)	Δ9	0	(18:0 to 18:1n-9)*
	Δ12	0	(18:1 to 18:2n-6)
	Δ15	0	(18:2n-6 to 18:3n-3)
	Δ6	28	(18:2n-6 to 18:3n-6)
	Δ6	37	(18:3n-3 to 18:4n-3)
	Δ5	0	(20:3n-6 to 20:4n-6)
pRSP3 ( <i>S. diclina</i> Δ5 desaturase)	Δ9	0	(18:0 to 18:1n-9)
	Δ12	0	(18:1 to 18:2n-6)
	Δ15	0	(18:2n-6 to 18:3n-3)
	Δ6	0	(18:2n-6 to 18:3n-6)
	Δ6	0	(18:3n-3 to 18:4n-3)
	Δ5	27	(20:3n-6 to 20:4n-6)

\*above endogenous Δ9 activity

Table 2  
Fatty Acid as a Percentage of Total Lipid Extracted from Yeast

Clone	18:1*	18:2 (n-3) Present	18:2 (n-6) Incorporated	18:3 (n-3) Produced	18:3 (n-6) Incorporated	18:3 (n-3) Produced	18:4 (n-3) Incorporated	18:4 (n-3) Produced	20:3(n-6) Incorporated	20:3(n-6) Produced	20:4 (n-6) Incorporated	20:4 (n-6) Produced
pYX242 (Control)	15.07	0	11.14	0	0	11.35	0	0	11.55	0		
pkSP1 ( $\Delta 6$ )	14.41	0	6.31	0	2.44	7.95	4.63	13.70	0			
pkSP3 ( $\Delta 5$ )	15.34	0.08	10.72	0	0	10.43	0	20.69	7.74			

50  $\mu$ M substrate added

\* 18:1 is an endogenous fatty acid in yeast

Key:

- 18:1 = Oleic acid
- 18:2 (n-6) = Linoleic acid
- 18:3 (n-3) =  $\alpha$ -Linolenic acid
- 18:3 (n-6) =  $\gamma$ -Linolenic acid
- 18:4 (n-3) = Stearidonic acid
- 20:3 (n-6) = Dihomo- $\gamma$ -linolenic acid
- 20:4 (n-6) = Arachidonic acid

Table 3

Fatty Acid as a Percentage of Total Lipid Extracted from Yeast

Clone	Plasmid in yeast (enzyme)	18:2 (n-6) Incorporated	18:3 (n-6) Produced	20:3 (n-6) Produced	% Conversion	18:3 (n-3) Incorporated	18:4 (n-3) Produced	20:4 (n-3) Produced	% Conversion
Control	pYX242 + pYES2	6.46	0	0	0	13.26	0	0	0
prSP5	prSP1 (Δ6) + pRAE73-A3 (human elongase)	4.62	1.95	0.8	37.3	7.00	2.47	1.20	34.39
prSP8	prSP1 (Δ6) + prPBP2 (M. alpina elongase)	4.08	2.31	0.63	41	5.93	2.01	0.85	32.53

50 μM substrate added

## Key:

18:2 (n-6) = Linoleic acid

18:3 (n-3) = α-Linolenic acid

18:3 (n-6) = γ-Linolenic acid

18:4 (n-3) = Stearidonic acid

20:3 (n-6) = Dihomo-γ-linolenic acid

20:4 (n-3) = Eicosatetraenoic acid

$$\% \text{ Conversion} = \frac{\% \text{ Product 1} + \% \text{ Product 2}}{[\% \text{ substrate} + \% \text{ Product 1} + \% \text{ Product 2}]}$$

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Table 4

Fatty Acid as a Percentage of Total Lipid Extracted from Yeast

Clone	Plasmid in yeast (enzyme)	18:3 (n-6) Incorporated	20:3 (n-6) Produced	20:4 (n-6) Produced	% Conversion	18:4 (n-3) Incorporated	20:4 (n-3) Produced	20:5 (n-3) Produced	% Conversion
Control	PYX242 + PYES2	8.17	0	0	0	5.61	0	0	0
pRSP7	pRSP3 ( $\Delta 5$ ) + pRAE73-A3 (human elongase)	6.25	2.30	1.63	38.6	4.12	1.98	1.56	46.2
pRSP10	pRSP3 ( $\Delta 5$ ) + pRPB2 (M. alpina elongase)	7.00	2.07	1.35	32.82	4.66	1.50	1.61	40.02

50  $\mu$ M substrate added

## Key:

- 18:3 (n-6) =  $\gamma$ -Linolenic acid  
 18:4 (n-3) = Stearidonic Acid  
 20:3 (n-6) = Dihomo- $\gamma$ -linolenic acid  
 20:4 (n-6) = Arachidonic Acid  
 20:4 (n-3) = Eicosatetraenoic acid  
 20:5 (n-3) = Eicosapentaenoic Acid

$$\% \text{ Conversion} = \frac{\% \text{ Product 1} + \% \text{ Product 2}}{\% \text{ substrate} + \% \text{ Product 1} + \% \text{ Product 2}}$$

**Table 5**

*Thraustochytrium aureum* (ATCC 34304) Desaturase Expression in Baker's Yeast

CLONE	TYPE OF ENZYME ACTIVITY	% CONVERSION OF SUBSTRATE
PRTA4 ( <i>T. aureum</i> Δ5 desaturase)	Δ9	0 (18:0 to 18:1n-9)*
	Δ12	0 (18:1 to 18:2n-6)
	Δ15	0 (18:2n-6 to 18:3n-3)
	Δ6	0 (18:2n-6 to 18:3n-6)
	Δ6	0 (18:3n-3 to 18:4n-3)
	Δ5	23.7 (20:3n-6 to 20:4n-6)
	Δ17	0 (20:4n-6 to 20:5n-3)
	Δ19	0 (22:4n-6 to 22:5n-3)
	Δ4	0 (22:4n-6 to 22:5n-6)
	Δ4	0 (22:5n-3 to 22:6n-3)

\*above endogenous Δ9 activity

Table 6

## Fatty Acid as a Percentage of Total Lipid Extracted from Yeast

50  $\mu$ M substrate added

\*18:1 is an endogenous fatty acid in yeast

Key:

- |                                   |   |
|-----------------------------------|---|
| Oleic acid                        | $\text{CH}_3(\text{CH}_2)_{7}\text{CH}=\text{CH}(\text{CH}_2)_{7}\text{COOH}$   |
| =Linoleic acid                    | $\text{CH}_3(\text{CH}_2)_{6}\text{CH}=\text{CH}(\text{CH}_2)_{4}\text{CH}=\text{CH}(\text{CH}_2)_{4}\text{COOH}$   |
| = $\alpha$ -Linolenic acid        | $\text{CH}_3(\text{CH}_2)_{6}\text{CH}=\text{CH}(\text{CH}_2)_{3}\text{CH}=\text{CH}(\text{CH}_2)_{3}\text{CH}=\text{CH}(\text{CH}_2)_{3}\text{COOH}$     |
| = $\gamma$ -Linolenic acid        | $\text{CH}_3(\text{CH}_2)_{6}\text{CH}=\text{CH}(\text{CH}_2)_{5}\text{CH}=\text{CH}(\text{CH}_2)_{5}\text{CH}=\text{CH}(\text{CH}_2)_{5}\text{COOH}$     |
| =Stearidonic acid                 | $\text{CH}_3(\text{CH}_2)_{16}\text{CH}=\text{CH}(\text{CH}_2)_{6}\text{CH}=\text{CH}(\text{CH}_2)_{6}\text{CH}=\text{CH}(\text{CH}_2)_{6}\text{COOH}$    |
| =Dihomo- $\gamma$ -linolenic acid | $\text{CH}_3(\text{CH}_2)_{16}\text{CH}=\text{CH}(\text{CH}_2)_{8}\text{CH}=\text{CH}(\text{CH}_2)_{8}\text{CH}=\text{CH}(\text{CH}_2)_{8}\text{COOH}$    |
| =Arachidonic acid                 | $\text{CH}_3(\text{CH}_2)_{16}\text{CH}=\text{CH}(\text{CH}_2)_{10}\text{CH}=\text{CH}(\text{CH}_2)_{10}\text{CH}=\text{CH}(\text{CH}_2)_{10}\text{COOH}$ |

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Table 7

Fatty Acid as a Percentage of Total Lipid Extracted from Yeast

Clone	20:3 Incorporated	20:4 Produced	22:4 Produced	Conversion to products
PYX242/ PYES2 (control)	41.98	0	0	0
PRTA4( $\Delta 5$ )/ PRAE73-A3 (human elongase)	15.59	4.2	6.28	16.7

100  $\mu$ M substrate added

\*18:1 is an endogenous fatty acid in yeast

Key:

 $\gamma$ -18:3 =  $\gamma$ -Linolenic acid20:3 = Dihomo- $\gamma$ -linolenic acid

20:4 = Arachidonic acid

22:4 = Adrenic acid

Fatty acid profiles of yeast containing pRAT-2c, pYX242, pRAT-2c/pRAE-73-A3, or pYX242/pYES2, grown in the presence of various fatty acids.

Table 9 Fatty acid profiles of yeast containing pRAT-1a, pYX242, pRAT-1a/pRAE-73-A3, or pYX242/pYES2, grown in the presence of various fatty acids.